Remarks

Reconsideration of this Application is respectfully requested.

Claims 1-112 are pending in the application, with claims 5, 48, 61, 85, 93, 95, 97, and110 being the independent claims.

Applicants gratefully acknowledge the withdrawal of rejections under 35 U.S.C. §§ 102 and 112, second paragraph.

Based on the following remarks, Applicants respectfully request that the Examiner reconsider all outstanding objections and rejections and that they be withdrawn.

Rejection under 35 U.S.C. § 112, first paragraph

The Examiner has withdrawn the allowability of claims 11, 15, 20-21, 45, 48, 63, 70, and 77, and rejected claims 1-9, 11, 15, 20, 21, 29, 42, 45, 48, 63, 70, 77, 93, and 94-110 under 35 U.S.C. § 112, first paragraph, because the specification, while being enabling for synthesizing and using compounds according to the present invention wherein the Q^+ is N (nitrogen), allegedly does not provide enablement for synthesizing and using compounds wherein the Q^+ moiety is S (sulfur) or O (oxygen). The Examiner further alleges that the specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to use the invention commensurate in scope with these claims. Applicants respectfully traverse this rejection.

Applicants note that claims 5-9, 29, 42, and 95-100 are directed to amine derivatives, i.e., to compounds where Q^+ is N. Thus, based on the Examiner's arguments, Applicants

assume the Examiner has inadvertently rejected claims 5-9, 29, 42, and 95-100 under 35 U.S.C. § 112, first paragraph, and request that the rejection be withdrawn.

The enablement requirement of § 112, first paragraph, ensures that one skilled in the art will be able to make and use the invention. Further, the test of enablement is not whether any experimentation is necessary, but whether, if experimentation is necessary, it is undue. M.P.E.P. § 2164.01. Furthermore, there is "no magical relation between the number of representative examples and the breadth of the claims; the number and variety of examples are irrelevant if the disclosure is 'enabling' and sets forth the 'best mode contemplated.'" *In re Borkowski*, 164 U.S.P.Q. 642 (C.C.P.A. 1970). A specification, in fact, need not contain a single working example. *Id.* 164 U.S.P.Q. at 645. Furthermore,

> [f]or a claimed genus, representative examples together with a statement applicable to the genus as a whole will ordinarily be sufficient if one skilled in the art (in view of level of skill, state of the art and the information in the specification) would expect the claimed genus could be used in that manner without undue experimentation. Proof of enablement will be required for other members of the claimed genus only where adequate reasons are advanced by the Examiner to establish that a person skilled in the art could not use the genus as a whole without undue experimentation.

M.P.E.P. § 2164.02.

Further, an applicant is not limited to the confines of the specification to provide the necessary information to enable the invention; there are two other sources of enabling information. *In re Howarth*, 654 F.2d 103, 105-6, 210 U.S.P.Q. 689, 692 (C.C.P.A. 1981). First, an applicant need not supply information that is well known in the art. *Genentech*, 108 F.3d at 1366, 42 U.S.P.Q.2d at 1005; *Howarth*, 654 F.2d at 105-6, 210 U.S.P.Q. at 692; *see also In re Brebner*, 455 F.2d 1402, 173 U.S.P.Q. 169 (C.C.P.A. 1972). "That which is

common and well known is as if it were written out in the patent and delineated in the drawings.'" *Howarth*, 654 F.2d at 106, 210 U.S.P.Q. at 692 (quoting *Webster Loom Co. v. Higgins* et al., 105 U.S. (15 Otto.) 580, 586 (1881)). Moreover, one of ordinary skill in the art is deemed to know not only what is considered well known in the art but also where to search for any needed starting materials. *Id.* Second, an applicant may incorporate by reference the necessary information by citing to certain types of documents that contain this information. *Id.*

The Examiner has the initial burden to establish a reasonable basis to question the enablement provided for the claimed invention. *In re Wright*, 27 U.S.P.Q.2d 1510 (Fed. Cir. 1993). Factors to be considered in determining whether pending claims would require undue experimentation have been articulated by the Court of Appeals for the Federal Circuit in *In re Wands*, 8U.S.P.Q.2d 1400 (Fed. Cir. 1988). They include:

> (1) the quantity of experimentation necessary, (2) the amount of direction or guidance presented, (3) the presence or absence of working examples, (4) the nature of the invention, (5) the state of the prior art, (6) the relative skill of those in the art, (7) the predictability or unpredictability of the art, and (8) the breadth of the claims.

Further,

> [i]t is improper to conclude that a disclosure is not enabling based on an analysis of only one of the above factors while ignoring one or more of the others. The examiner's analysis must consider all the evidence related to each of these factors, and any conclusion of enablement must be based on the evidence as a whole.

M.P.E.P. § 2164.01(a).

In this case, the Examiner has not set forth evidence to demonstrate that one skilled in the art would find the specification nonenabling in light of its discussion and

exemplification. Additionally, the Examiner has not provided the Applicants with an assessment of enablement under the "standard of reasonableness" to which they are entitled, given the nature of the invention and the state of the art.

> The determination of what constitutes undue experimentation in a given case requires the application of a standard of reasonableness, having due regard for the nature of the invention and the state of the art. [citations omitted] The test is not merely quantitative, since a considerable amount of experimentation is permissible, if it is merely routine, or if the specification in question provides a reasonable amount of guidance with respect to the direction in which the experimentation should proceed . . ..

In re Jackson, 217 U.S.P.Q. 804 (Bd. App. 1982, cited with approval in *Wands*, 8 U.S.P.Q.2d at 1404).

Even when "unpredictability" in a field such as chemistry may create reasonable doubt as to the accuracy of a broad statement supporting enablement, and even when the statement is, on its face, contrary to generally accepted scientific principles, the Court of Customs and Patent Appeals has clearly stated that

> [I]t is incumbent upon the Patent Office, whenever a rejection on this basis is made, to explain why it doubts the truth or accuracy of any statement in a supporting disclosure and to back up assertions of its own with acceptable evidence or reasoning which is inconsistent with the contested statement.

In re Marzocchi, 169 U.S.P.Q. 367 (C.C.P.A. 1967).

The reasons behind the Examiner's rejection are as follows:

> There are no examples wherein Applicants have either synthesized compounds wherein Q^+ is either S or O, and further wherein Applicants used these compounds for the delivery of macromolecules into cells.

At page 3, lines 3-5, of the Office Action.

> [D]ue to the differences in the chemical properties between Oxygen, Sulfur, and Nitrogen, one of skill in the art would not accept on its face that the synthetic route for the compounds according to the invention wherein Q^+ is N is readily amenable for the synthesis of compounds wherein the Q^+ moiety is either O or S.

At page 3, lines 5-9, of the Office Action.

> Based upon the lack of disclosure with regards to the synthetic steps required to synthesize the full breadth of compounds encompassed by the Markush structure recited in claims 1, 93, and 110, and the lack of working examples wherein Applicants have successfully synthesized compounds wherein Q^+ is either S or O, and wherein Applicants have used said compounds for delivery of macromolecules in the cells, one of ordinary skill in the art would have to resort to trial and error experimentation in order to practice the full scope of the claimed invention.

At page 3, lines 9-15, of the Office Action.

> The quantity of experimentation required to practice the invention as claimed would require determining the synthetic routes for isolating the full scope of the compounds encompassed by the instant claims, and further determining if said compounds are suitable as transfection agents.

At page 3, lines 16-19, of the Office Action.

> The specification as filed provides only guidelines with regards to substituted amine compounds according to the general formula (A).

At page 3, lines 19-20, of the Office Action.

> The deficiencies in the specification would constitute undue experimentation since the steps required for the chemical preparation of compounds according to the general formula (A) wherein Q^+ is either S or O, must be determined without instructions from the specification before one is enabled to practice the claimed invention.

At page 3, line 20 through page 4, line 2, of the Office Action.

Considering the factors articulated by the Federal Circuit in *Wands*, it is respectfully submitted that Applicants' invention does not reasonably require undue experimentation.

Factor No. 1, quantity of experimentation. The Examiner did not seek to quantify the amount of experimentation, but rather noted in a conclusory fashion that undue experimentation would be required since the synthetic steps for preparing compounds of formula (A) wherein Q^+ is either S or O must be determined without instructions from the specification.

Factor No. 2, the amount of direction or guidance presented. The Examiner alleges that guidance is presented only with regard to substituted amine compounds of general formula (A). However, the specification teaches the degree of substitution in compounds of formula (A) where Q^+ is S or O at page 50, lines 7-8, as follows:

> It would be obvious for a skilled person that when Q is O or S, the number of substituents should be according to their valency.

Further, the specification teaches that "the reaction schemes provide a general method for preparing a variety of compounds according to the present invention" and that "alternate methods and reagents other than those specifically detailed herein can be employed or readily adapted to produce compounds of the invention." At page 71, lines 10-14 of the specification. Furthermore, the specification teaches various, structurally different compounds that can be useful in delivering macromolecules and other compounds into cells at pages 2-5 of the specification. The specification also teaches various methods for incorporating cationic lipids into lipid aggregates at page 71, lines 15-29, of the specification. Furthermore, the specification teaches that methods for delivering macromolecules and other compounds into cells are well-known in the art and particularly

demonstrates the delivery of plasmid-DNA into several different type of cells in examples 7 and 8, at page 72, line 22 through page 75, line 24 of the specification.

It is known in the art that -NH-, -S- and -O- are classical bioisosters and, thus, exhibit similar chemical and physical properties, and produce broadly similar biological properties. Thornber, C. W., *Rev. Chem. Soc. 8*:563-580 (1979). Therefore, in view of the general teaching of the present application and, specifically, the teaching regarding substituted amine compounds (see, e.g., Schemes 1-5 of the specification), the methods for obtaining substituted sulfur and oxygen compounds according to the invention and using them according to the invention would be reasonably apparent to a person of skill in the art. A copy of Thornber is attached as Appendix A.

The specification has incorporated by reference U.S. Patent No. 5,674,908 ("the '908 patent") to Haces *et al.* that describes general reaction schemes for synthesizing dicationic and polycationic sulfonium lipids (see Scheme 3 and Scheme 4 at col. 12). A copy of the '908 patent has been provided as document AE2 along with an Information Disclosure Statement filed on March 9, 2000. Furthermore, dithiol compounds suitable for starting materials are commercially available or can be prepared by methods known in the art. For example, the dithiol starting material corresponding to jeffamine in Scheme 1 at page 60 of the present application, i.e., 1,4-butanedithiol, is commercially available by, e.g., Acros-USA.

Since the bivalent atoms S and O exhibit similar chemical and physical properties, it would be reasonable to expect a person skilled in the art could prepare oxygen derivatives according to the present invention having the teaching of the instant specification and the '908 patent. Suitable diol starting materials are commercially available or can be prepared

by methods known in the art. In particular, the diol starting material corresponding to jeffamine in Scheme 1 at page 60 of the present application, i.e., 1,4-butanediol, is also commercially available by, e.g., Acros-USA.

In view of the above, the Examiner has not provided any factual basis on which it can be concluded that one skilled in the art would not be able to prepare compounds of formula (A) of the present invention wherein Q^+ is S or O and use them for delivering macromolecules or other compounds into cells.

Factor No. 3, the presence or absence of working examples. The specification provides a number of working examples on substituted amine compounds of formula (A) as admitted by the Examiner. Also the delivery of plasmid-DNA into different cells using those compounds has been demonstrated (see *Factor No. 2*).

As stated above, a specification need not contain a single working example, and representative examples together with a statement applicable to the genus as a whole will be sufficient if one skilled in the art would expect the claimed genus could be used in that manner without undue experimentation. It is respectfully submitted that the Examiner did not provide adequate reasons to establish that a person skilled in the art could not make and use compounds of the present invention other than the amine derivatives of formula (A).

Factor No. 4, the nature of the invention. The Examiner did not seek to determine the nature of the invention.

Factor No. 5, the state of the art. The Examiner did not address this issue, but merely concluded that based on chemical differences between N, S, and O, a person skilled in the art would not accept that the synthetic routes for amine derivatives of formula (A) are readily amenable for sulfur and oxygen derivatives. However, the Examiner did not provide

any evidence or compelling rationale why the art of preparing lipids having oxygen or sulfur as a cation instead of nitrogen, and their use in delivering macromolecules and other compounds into cells is so inherently complex as to permit no generalizations beyond nitrogen as the Q^+ moiety.

Factor No. 6, the relative skill of those in the art. The Examiner did not address this issue. Given the state of the art and the source of publications and patents in the art, it is clear that the skill level in the art of lipids and their use for delivery of macromolecules and other compounds into cells is high, typically that of a Ph.D. level worker.

Factor No. 7, the breadth of the claims. The Examiner did not provide any evidence or compelling rationale to support the finding that the invention disclosed by Applicants could not be practiced to the full scope of the pending claims.

In conclusion, this *prima facie* § 112 rejection is improper because it fails to comply with the law as enunciated in *In re Wands* and fails to provide a rational basis for the allegation of "undue experimentation."

In view of the above, reconsideration and withdrawal of the rejection of claims 1-9, 11, 15, 20, 21, 29, 42, 45, 48, 63, 70, 77, 93, and 94-110 under 35 U.S.C. § 112, first paragraph, are respectfully requested.

Objections

The Examiner has objected to claims 10, 12-14, 16-19, 22-28, 43-44, 46-47, 49-62, 64-69, 71-76, 78-92, and 111-112 as being dependent upon a rejected base claim, but would be allowable if rewritten in independent form including all of the limitations of the base claim and any intervening claims.

In view of the above, Applicants believe the objection has been rendered moot. Accordingly, reconsideration and withdrawal of the objection to claims 10, 12-14, 16-19, 22-28, 43-44, 46-47, 49-62, 64-69, 71-76, 78-92, and 111-112 are respectfully requested.

Conclusion

All of the stated grounds of objection and rejection have been properly traversed, accommodated, or rendered moot. Applicants therefore respectfully request that the Examiner reconsider all presently outstanding objections and rejections and that they be withdrawn. Applicants request the entry of this Amendment, the Examiner's reconsideration and reexamination of the application, and the timely allowance of the pending claims.

It is believed that a full and complete reply has been made to the outstanding Office Action and, as such, the present application is in condition for allowance. If the Examiner believes, for any reason, that personal communication will expedite prosecution of this application, the Examiner is invited to telephone the undersigned at the number provided.

Respectfully submitted,

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Isosterism and Molecular Modification in Drug Design

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1 Introduction

The idea of isosterism goes back to Langmuir[1] in 1919. At that time the word isosterism was used to describe the similarity of molecules or ions which have the same number of atoms and valence electrons *e.g.* O^{2-}, F^-, Ne. Clearly only those isosteres with the same nett charge show similar chemical and physical properties. Grimm[2] enunciated his hydride displacement law to describe the similarity between groups which have the same number of valence electrons but different numbers of atoms. For example some similarities are present in the sequence: CH_3, NH_2, OH, Hal.

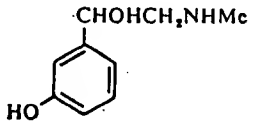
Grimm's hydride displacement law points out some similarities of size in groupings based on elements in the same row of the periodic table. Other similarities to be found in the periodic table are within the groups, where chemical reactivities are similar but with electronegativity decreasing as atomic weight increases and lipophilicity and polarizability increasing with the size of the atom. Other relationships exist in diagonal lines across the periodic table where atoms of similar electronegativity such as nitrogen and sulphur, oxygen and chlorine are found.

In trying to relate biological properties to the physical and chemical properties of atoms, groups, or molecules, many physical and chemical parameters may be involved and the simple relationships mentioned above are clearly inadequate for this purpose. Friedman[3] introduced the term 'bioisosterism' to describe the phenomenon in which compounds which are related in structure have similar or antagonistic properties. The use of the word isosterism has clearly outgrown its original meaning when used in medicinal chemistry and a loose flexible definition could be adopted such as: 'Bioisosteres are groups or molecules which have chemical and physical similarities producing broadly similar biological properties'.

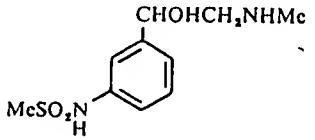
The term non-classical isosterism is also used interchangeably with bioisosterism, particularly in connection with isosteres which do not have the same number of atoms but do produce a similarity in some key parameter of importance in

[1] I. Langmuir, *J. Amer. Chem. Soc.*, 1919, 41, 868, 1543.
[2] H. G. Grimm, *Z. Elektrochem.*, 1925, 31, 474; 1928, 34, 430; 1934, 47, 53, 594.
[3] H. L. Friedman, 'Influence of Isosteric Replacements upon Biological Activity', National Academy of Sciences—National Research Council Publication No. 206. Washington D.C., 1951, p. 295.

that series. For example[4] the two β-adrenergic stimulants compounds (1) and (2) have similar activity.



(1) pK_a 9.6



(2) pK_a 9.1

The concept of bioisosterism has been described in reviews by Burger,[5a] Schatz,[5b] Foye,[6] Korolkovas,[7] Ariens,[8] and Hansch.[9] This present review collates and extends the earlier observations with more recent reports from the literature and suggests new techniques for exploiting the concept.

The 'classical' isosteres as defined by Burger[5] and Korolkovas[7] are given in Table 1.

Table 1

1) *Univalent atoms and groups*

F	OH	NH_2	Me	Cl
	SH	PH_2		
	I	Bu^t		
	Br	Pr^i		

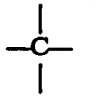
2) *Bivalent atoms and groups*

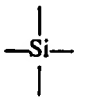
O	S	Se	CH_2	—N(H)—
CO_2R	COSR	$COCH_2R$	CONHR	

3) *Tervalent atoms and groups*

—N=	—CH=
—P=	—As=

4) *Quadrivalent atoms*





5) *Ring equivalents*

—CH=CH—	S	*e.g.* benzene: thiophen
=C—	=N—	*e.g.* benzene: pyridine
H		

—O— —S— —CH_2— —NH—

[4] A. A. Larson and P. M. Lish, *Nature*, 1964, 203, 1283.
[5a] A. Burger in 'Medicinal Chemistry' 3rd Edn., ed. A. Burger, Wiley-Interscience, New York, 1970.
[5b] V. B. Schatz in 'Medicinal Chemistry' 2nd Edn., ed. A. Burger, Wiley-Interscience, New York, 1960.
[6] W. O. Foye, 'Principles of Medicinal Chemistry', Lea and Febiger, Philadelphia, 1970.
[7] A. Korolkovas, 'Essentials of Molecular Pharmacology: Background for Drug Design', Wiley, 1970.
[8] E. J. Ariens in 'Drug Design', ed. E. J. Ariens, Academic Press, New York, 1971, Vol. 1.
[9] C. Hansch, *Intra-Science Chem. Rep.*, 1974, 8, 17.

2 Bioisosterism in Molecular Modification

In the process of developing a lead compound, an antagonist to a known agonist, or an anti-metabolite from a known substrate, a large number of systematic molecular modifications will be made. The modern concept of bioisosterism can be an aid to the design of such modifications. In making a bioisosteric replacement the following parameters of the group being changed could be considered:

(*a*) Size.

(*b*) Shape (bond angles, hybridization).

(*c*) Electronic distribution (polarizability, inductive effects, charge, dipoles).

(*d*) Lipid solubility.

(*e*) Water solubility.

(*f*) pK_a.

(*g*) Chemical reactivity (including likelihood of metabolism).

(*h*) Hydrogen bonding capacity.

It is unlikely that any bioisosteric replacement will leave all these parameters undisturbed. The extent to which the replacement is useful will depend upon which of these parameters is important and which ones the bioisostere can best mimic.

The element of a molecule being modified may have one or more of the following roles.

(i) *Structural.* If the moiety has a structural role in holding other functionalities in a particular geometry, parameters such as size and bond angle will be important. The moiety may be buried deep in the molecule and have little contact with the external medium.

(ii) *Receptor interactions.* If the moiety to be replaced is concerned with a specific interaction with a receptor or enzyme its size, shape, electronic properties, pK_a, chemical reactivity, and hydrogen bonding will be the important parameters.

(iii) *Pharmacokinetics.* The moiety to be replaced may be necessary for the absorption, transport, and excretion of the compound. In this case lipophilicity, hydrophilicity, hydrogen bonding, and pK_a are likely to be important.

(iv) *Metabolism.* The moiety may be involved in blocking or aiding metabolism. In this case chemical reactivity will be an important parameter. For example chloro and methyl substituents on a benzene ring may be interchangeable for certain purposes but the toluene derivative can be metabolized to a benzoic acid and may therefore have a shorter half-life or unexpected side effects.

Usually one will not know which role(s) the various parts of the molecule play(s) in its action and this determination will be part of the structure–activity study. However, from the simple considerations listed above it is clear that:–

(A) A given molecular modification may allow some, but probably not all of the parameters (*a*)—(*h*) to be kept the same.

(B) Whether the same or a different biological activity results from the replacement will be governed by the role(s) which that moiety fulfils in the molecule and whether parameters affecting that role have been disturbed.

(C) From (A) and (B) it follows that what proves to be a good bioisosteric replacement in one series of compounds will not necessarily be useful in another.

Completely identical properties are rarely sought and will in any case be difficult if not impossible to achieve. What we are more likely to be seeking is a subtle change in the molecule which will leave some properties the same and some different in order to improve potency, selectivity, absorption, duration, and toxicity. Bioisosteric replacements allow molecular modifications, in which the number of variables changed are limited. Ariens[8] and Korolkovas[7] have tried to introduce the idea of partial bioisosteric groups as those which turn an agonist into an antagonist. Although their lists of groups may be suggestive to the drug designer, the idea is probably incorrect because of the statement (C) above. An 'antagonist' group in one molecule will only antagonize a similar 'agonist' group in another molecule if the agonist groups in both series are performing the same function. If an isosteric replacement results in a molecule which has some properties similar to the parent molecule but some important property has changed, it may be possible to compensate for this undesirable change by modifications elsewhere in the molecule. For example a molecular modification may reduce the lipid solubility of the molecule thereby affecting its absorption, transport, and apparent potency. Optimum activity may be regained by inserting lipophilic groups into the molecule at some sterically undemanding site. Consequently the best compounds in this parallel series of isosteres, such as for example furans and thiophens, are likely to have different substituent patterns.

3 The Mathematical Formulation

The arguments used above can be expressed in the mathematical form used by Hansch[10] for the case where a simple substituent is being varied, for example on a benzene ring. If the potency of a drug is a function of several parameters of the substituent then:

$$\log \frac{1}{c} = A(\pi) + B(\sigma) + C(E_s)$$

where Hansch's π value is used for the lipophilic character, Hammett's σ value for the electronic property, Taft's steric parameter to denote the size of the group and c is the concentration of drug required to achieve a given effect.

If such a relationship were found for a drug series in which the constants B and C were zero then the potency would be a function of π only. In this context groups would be bioisosteric if they have similar π values independent of their

[10] C. Hansch, *Accounts Chem. Res.*, 1969, 2, 232.

σ and E_s values. If however the three constants A, B, and C are all significant a much more limited range of equivalent groups will be available.

If a series of compounds has more than one property, as is usual, then more than one equation will be needed to describe the effects of changing the substituent:

$$\log \frac{1}{c} = A(\pi) + B(\sigma) + C(E_s)$$

Desired activity

$$\log \frac{1}{c} = D(\pi) + E(\sigma) + F(E_s)$$

Side effects

Clearly if $A = D$, $B = E$, and $C = F$, *etc.*, no selectivity can be found within this limited series. If however $C \ll F$ then for the desired activity E_s is not important and π and σ may be optimized while reducing the value of E_s, thereby reducing the side effects. This phenomenon of increasing selectivity by bioisosteric replacement relies upon the fact that some desirable properties in the molecule can be retained when unimportant parameters can be varied. An unimportant parameter for the biological activity desired *may* be a key parameter in the side effect.

Thus bioisosteric replacements are useful in searching for potency, selectivity, absorption, and duration. Following the Hansch treatment one could produce a modern definition of bioisosterism based upon measurable parameters such as π, σ, E_s, hydrogen bonding properties, pK_a, *etc.*, and Hansch[9] has used the term 'isolipophilic' for groups with the same π value.

Table 2 shows some functional groups with similar electron-withdrawing properties. If electronic effects alone influence the biological activity in a series of drugs then these groups would be equivalent. If, however, the lipophilicity and steric factors are important then absolute identity cannot be achieved.

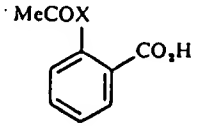
Table 2

Functional Group	σ_m	π	E_s
F	0.34	0.14	0.78
Cl	0.37	0.71	9.27
Br	0.39	0.86	0.08
I	0.35	1.12	−0.16
CF_3	0.43	0.88	−1.16
SCF_3	0.40	1.44	
COMe	0.31	−0.55	
CHO	0.36	−0.65	
CO_2Me	0.32	−0.01	
CH=CH—NO_2	0.32	0.11	

Extensive tables of σ, π, and E_s values are now available.[11] These can be used to gain a more quantitative idea of some aspects of isosterism using the better known functional groups.

4 Chemical Reactivity

Biological effects are generally produced by 'weak' interactions between the drug and the receptor but covalent bonding does occasionally play a part. A series of aspirin isosteres (3) was reported in 1975.[12] The nitrogen, sulphur, and carbon



(3) X = O, NH, S, or CH_2

isosteres were all totally inactive despite the classical purity of the replacements tried. Now that it is known that aspirin is an acetylating agent for prostaglandin synthetase this result is more readily understood.[13] The agents are widely different in their ability to act as acylating agents unless other substantial modifications are made in the molecules.

5 Non-classical Isosteres: Some Further Points

In considering bioisosterism in its widest sense it should be noted that similar effects in two functional groups need not imply atom upon atom overlap. Edwards[14] has pointed out that a common enzyme or receptor interaction involves hydrogen bonding to a carbonyl group. Strong hydrogen bonds may be formed to the carbonyl oxygen by hydrogen atoms within a cone having an angle of about 60° at its apex. Two molecules RXH and RAXH, where A is an additional atom, may be able to bind to the active site without identical positioning of the X or H. In addition the conformational mobility in both the drug and the receptor molecule will allow essentially similar binding of two drugs without the need to consider that the binding groups on the drugs are positioned in space in an identical manner.

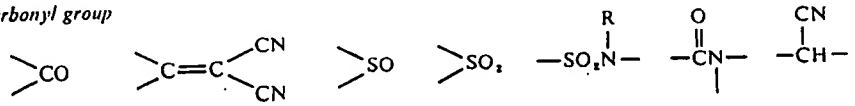
Examples of Non-classical Isosteres.—The list shown in Table 3 is drawn from earlier reviews[5–9] and from the examples given in Table 4 at the end of this

[11] Tables of substituent constants can be found in the following papers. C Hansch, S. D. Rockwell, P Y. C. Jow, A. Leo, and E. E. Steller, *J. Med. Chem.*, 1977, 20, 304; J. G. Topliss, *J. Med. Chem.*, 1972, 15, 1006, and 1977, 20, 463; C. Hansch, A. Leo, S. H. Unger, Ki Hwan Kim, D. Nikaitoni, and E. J. Lien, *J Med. Chem.*, 1973, 16, 1207.
[12] L. Thompkins and K. H. Lee, *J. Pharm. Sci*, 1975. 64, 760.
[13] G. J. Roth, N Stanford, and P. W. Majerus, *Proc. Nat. Acad. Sci, U.S.A.*, 1975, 72, 3073.
[14] P. N. Edwards, I.C.I. Pharmaceuticals Division, personal communication.

review. In addition a few proposals[15–17] which have not yet been realized in medicinal chemical work are included.

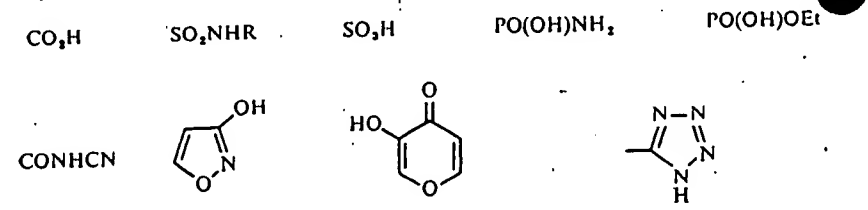
Table 3

Carbonyl group



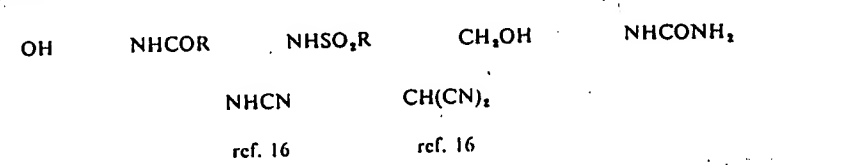
ref. 15

Carboxylic acid group



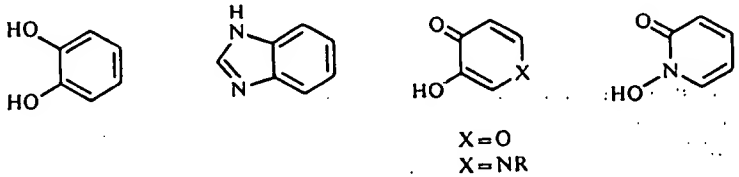
ref. 16

Hydroxy-group



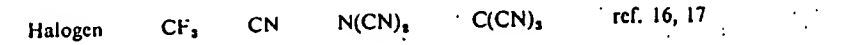
ref. 16 ref. 16

Catechol



X=O
X=NR

Halogen

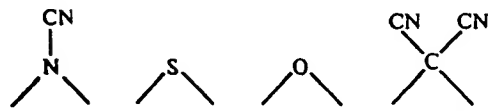


ref. 16, 17

[15] K. Wallenfels, K. Friedrich, J. Rieser, W. Ertel, and H. K. Thieme, *Angew. Chem. Internat. Edn.*, 1976, 15, 261.
[16] H. von Kohler, B. Eichler, and R. Salewski, *Z. anorg. Chem.*, 1970, 379, 183, also includes other possibilities in the sulphur and phosphorus and nitro acid series.
[17] K. von Wallenfels, *Chimia*, 1966, 20, 303.

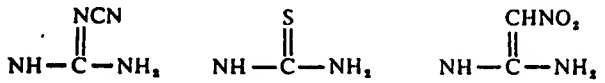
Table 3 *continued*

Thioether



ref. 16

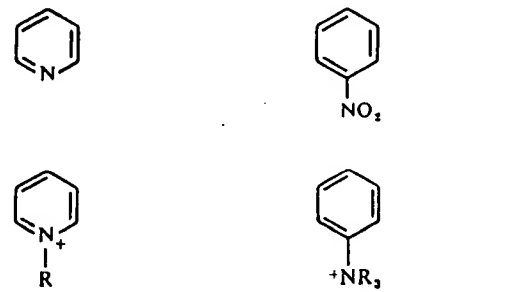
Thiourea



Azomethine

ref. 17

Pyridine



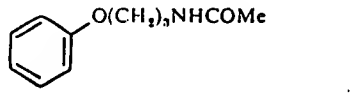
Spacer groups

In addition ring-opened forms of molecules may be considered to be isosteric with the corresponding ring-closed forms although the conformation of the *seco* form will be unlike the parent molecule. However, if in ring opening an atom is removed a conformation similar to the parent molecule may be possible.

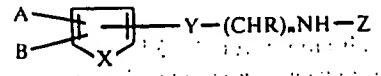
6 Substructure Searching and Bioisosterism

Although the classical Hansch approach is used largely for optimization within a series, molecular modifications based on bioisosterism principles can generate new series or even develop new leads if an agonist is used as the starting point for the design of an antagonist. One aid to this process is the use of a compound collection and computer techniques for doing substructure searches, *e.g.* the

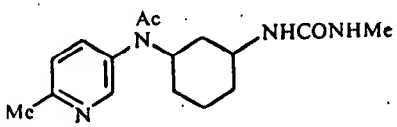
Crossbow suite of programmes.[18] For example suppose that random screening has turned up the lead (4). One may consider bioisosteric replacements for the ring, the oxygen, the polymethylene chain, or the amidic moiety, and design a substructure search for compounds of type (5). A vast number of permutations are possible and from these compounds may be available for tests which result in new leads which have properties worth exploiting, such as perhaps (6).



(4)



(5) X = CH=CH, CH=N, S, O, or NR
Y = O, S, SO, SO_2, Se, NCN, or NCOR
n = 2, 3, or 4
R = H or alkyl, including forming a ring
Z = COR, CO_2R, SOR, SO_2R, or CONHR
A = B = defined substituents

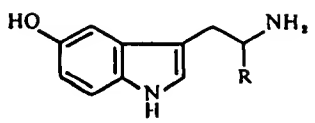


(6)

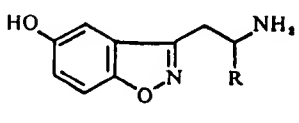
Examples.—The literature of medicinal chemistry is rich in examples of the use of the concept of bioisosterism and the reader is referred to the reviews mentioned[5–8] and the references quoted therein for examples reported before 1970. There follows a brief discussion of bioisosteres of some indole–amines which has some useful lessons, and Table 4 lists examples culled from the literature since 1970. Only the structures are given in this Table as an illustration of the kinds of change which have been useful. The reader is referred to the original papers for the full details of biological activity and selectivity. The list is not comprehensive but represents some uses of more novel non-classical types. Rudinger[19] has reviewed isosteric replacements in the field of peptide chemistry up to 1971 and some further discussions[20] have been published recently.

Indole-amines.—Campaigne[21] has studied and reviewed the work on bioisosteres of 5-hydroxytryptamine (7) and one or two details of the work are instructive. Whereas (8) was inactive as an agonist or antagonist on the rat uterus preparation, the corresponding tryptophan analogue (9) had weak activity as an enzyme inhibitor for 5-hydroxytryptamine decarboxylase.[22] This type of bioisostere

[18] E. E. Townsley and W. A. Warr, 'Chemical and Biological Data; An Integrated On-Line Approach' in 'Retrieval of Medicinal Chemical Information', ed. Howe, Milne, and Pennell (A. C. S. Symposium Series No. 84), American Chemical Society, Washington D.C.
[19] J. Rudinger, in ref. 8, Vol. II, Chapter 9.
[20] Further discussion of peptide backbone replacement is found in ref. 19 and W. Soudyn and I. van Wijngaarden, in 'Biological Activity and Chemical Structure', ed. J. A. Keverling Buisman, Elsevier, Holland, 1977; a peptide link isostere —CH_2—S— has been reported by J. A. Yankeelov, Kam-Fook Fok, and D. J. Carothers, *J. Org. Chem.*, 1978, 43, 1623.
[21] E. Campaigne, R. P. Maichel, and T. R. Bosin, Medicinal Chemistry, Specialist Contributions, 3rd International Symposium, 1972, Butterworths, 1973, p. 65.
[22] M. Pigini, M. Gianella, F. Gualtieri, C. Melchiorne, P. Bolle, and L. Angelucci, *European J. Med. Chem.*, 1975, 10, 29, 33.

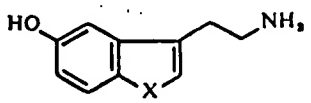


(7) R = H (5-hydroxytryptamine)
(10) R = CO_2H (5-hydroxytryptophan)



(8) R = H
(9) R = CO_2H

loses all affinity for the 5-hydroxytryptamine (5-HT) receptor but retains it in part for an enzyme system. Similarly, in the series of compounds 5-HT, (11), (12), and (13) activity has been measured against the rat fundic strip preparation and on the enzyme caeruloplasmin.[23] Whereas 5-HT is a substrate for the enzyme, compound (11) inhibited caeruloplasmin's oxidation of 5-HT and noradrenaline.



	Rat Fundic Strip		
	X	*Intrinsic activity*	PD_2
5-HT	NH	1	7.6
(11)	CH_2	0.96	5.6
(12)	O	0.84	4.6
(13)	S	1.08	6.1

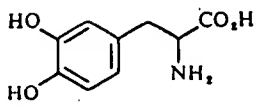
Compound (12) inhibits only 5-HT oxidation and compound (13) was inactive as a substrate or an antagonist. This would appear to demonstrate that for the enzyme system the imino grouping at the 1-position of the ring is essential.

On the rat fundic strip, however, all the analogues have full agonist activity though with reduced potency, demonstrating that the 5-HT receptor has a greater tolerance for loss of the imino nitrogen. These simple experiments demonstrate the role of bioisosteric replacements in exploring selectivity between different receptors and enzymes.

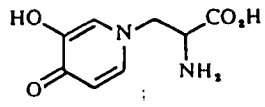
[23] B. C. Barrass, D. B. Goult, R. M. Pinder, and M. Sheels, *Biochem. Pharmacol.*, 1973, 22, 2891.

Table 4 *Some recent examples of bioisosterism*

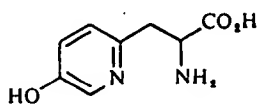
Dihydroxyphenylalanine analogues



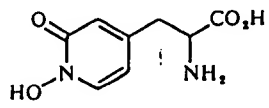
Dopa



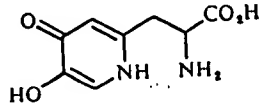
Mimosine ref. 24



ref. 25

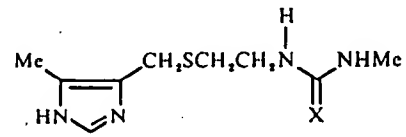


ref. 26

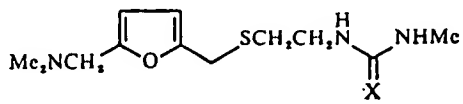


ref. 27

Histamine H-2 antagonists



X = S or NCN ref. 28

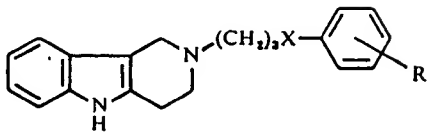


X = NCN or $CHNO_2$ ref. 29

[24] H. Haguchi, *Mol. Pharmacol.*, 1977, 13, 362.
[25] A natural product from *Streptomyces* species, S. Inoue, T. Shamura, T. Tsurvoka, Y Ogawa, H. Watanabe, J. Yoshidea, and T. Nuda, *Chem. and Pharm. Bull. (Japan)*, 1975, 23, 2669.
[26] Synthesized as a mimosine analogue, R. N. L. Harris and R. Teitei, *Austral. J. Chem.*, 1977, 30, 649.
[27] S. J. Norton and E. Sanders, *J. Med. Chem.*, 1967, 10, 961.
[28] R. W. Brimblecombe, W. A. M. Duncan, C. J. Durant, J. C. Emmett, C. R. Gannelin, and M. E. Parsons, *J. Int. Med. Res.* 1975, 3, 86. See also Sulphur–methylene isosterism in the development of metiamide, J. W. Black, G. J. Durant, J. C. Emmett, and C. R. Gannelin, *Nature*, 1974, 248, 65, and C. R. Gannellin, *J. Appl. Chem. Biotechnol.*, 1978, 28, 183.
[29] Allen and Hanbury, U.S.P. 4 128 658.

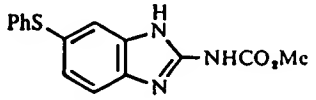
Table 4 *continued*

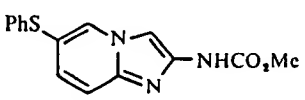
Neuroleptics



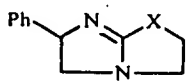
X = C(=O) or CH—CN ref. 30

Anthelmintics



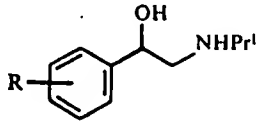


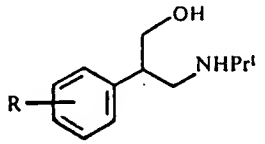
ref. 31



X = S or Se ref. 32

β-Adrenergic blockers

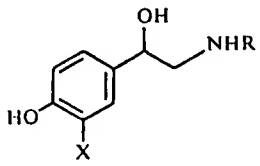




ref. 33

[30] Boehringer, Sohn C. H., U.S.P. 4 085 216.
[31] H. Fisher and M. Lusi, *J. Med. Chem.*, 1972, 15, 982; R. J. Bochis, R A. Dybas, P. Eskola, P. Kulsa, B. O. Linn, A. Lusi, E. Mutzner, J. Milkowski, H. Mrozik, L. E. Olen, L. H. Peterson, R L. Tolman, A. F. Wagner, F. S. Waksmunski, J. R. Egerton, and D. A. Osteind, *J. Med. Chem.*, 1978, 21, 235.
[32] R. N. Hanson, R. N. Giese, M. A. Davis, and S. M. Costello, *J. Med. Chem*, 1978, 21, 496.
[33] T. Jen, J. S. Frazee, M. S. Schwartz, K. F. Erhard, C. Kaiser, D. F. Colella, and J. R. Wardell, *J. Med. Chem.*, 1977, 20, 1263.

β-Adrenergic stimulants

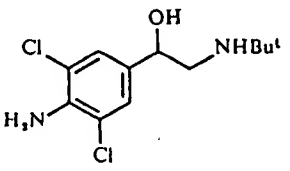


R = Me, X = OH Adrenaline

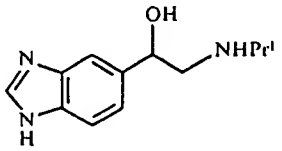
R = Bu^t, X = CH_2OH
Salbutamol ref. 34

R = Bu^t, X = $NHCONH_2$
Carbuterol ref. 35

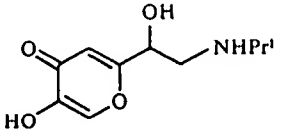
R = Pr^i, X = $NHSO_2Me$
Soterenol ref. 36



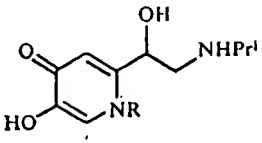
Clenbuterol ref. 37



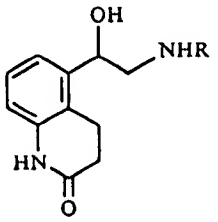
ref. 38



ref. 39



ref. 39

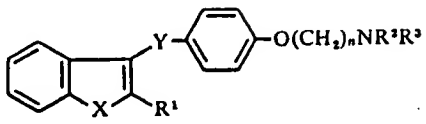


ref. 40

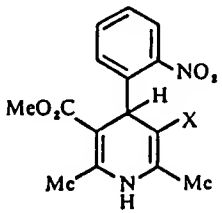
[34] D. Hartley, D. Jack, L. H. Lunts, and A. C. Ritchie, *Nature*, 1968, 219, 861; D. T. Collin, D. Hartley, D. Jack, L. H. C. Lunts, J. C. Press, A. C. Ritchie, and P. Toon, *J. Med. Chem.*, 1970, 13, 674.
[35] C. Kaiser, *J. Med. Chem.*, 1974, 17, 49.
[36] A. A. Larsen, W. A. Gould, H. R. Roth, W. T. Comer, R. H. Uloth, K. W. Dungan, and P. M. Lish, *J. Med. Chem.*, 1967, 10, 462.
[37] J. Keck, G. Kruger, K. Noll, and H. Machleidt, *Arzneimittelforsch.*, 1972, 22, 861.
[38] C. D. Arnett, J. Wright, and N. Zenker, *J. Med. Chem.*, 1978, 21, 72.
[39] H. W. R. Williams, *Canad. J. Chem.*, 1976, 54, 3377.
[40] S. Yoshizaki, K. Tarimura, S. Tamada, Y. Yabuuchi, and K. Nakagawa, *J. Med. Chem.*, 1976, 19, 1138.

Table 4 *continued*

Vasodilators

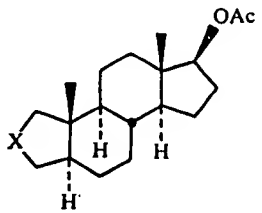


X = O or S, Y = SO_2 ref. 41
X = O, Y = CO ref. 42
X = S, Y = CO ref. 43



X = CO_2Me ref. 44
X = SO_2Me ref. 45

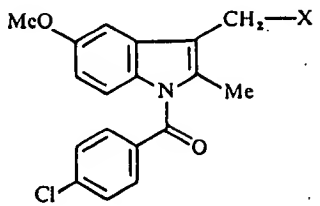
Androgens

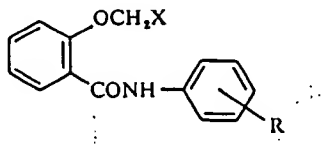


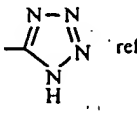
X = S or NCN ref. 46

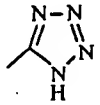
[41] SmithKline Corp., U.S.P. 4 117 128.
[42] E. M. Vaughan Williams and P. Polster, *European J. Pharmacol.*, 1974, 25, 241; *Unlisted Drugs.*, 1971, 23, (8), 110.
[43] N. Claeys, C. Goldenberg, R. Wandestrick, E. Devay, M. Descamps, G. Delaunois, J. Bauthier, and R. Charlier, *Chim. Ther.*, 1972, 7, 377.
[44] F. Bossert, and W. Vater, *Naturwiss.*, 1971, 58, 578; *Drugs of Today*, 1975, 11, 154.
[45] Ciba-Geigy B.P. 1 464 324.
[46] W.-H Chiu, T. H. Klein, and M. E. Wolff, *J. Med. Chem.*, 1979. 22, 119.

Anti-inflammatory



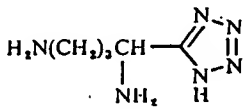


X = CO_2H or  ref. 49

X =  ref. 47

X = CO_2H ref. 48

Ornithine decarboxylase inhibitor



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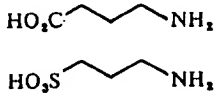
Gabergic agents



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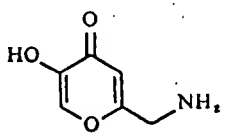

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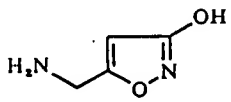
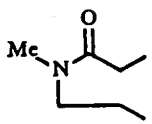


ref. 52

[47] P. F. Juby and T. W. Hudyma, *J. Med. Chem.*, 1969, 12, 396.
[48] T. Y. Shen, R. L. Ellis, T. B. Windholz, A. R. Matzuk, A. Rosegay, S. Lucas, B. E. Witzel, C. H. Stammer, A. N. Wilson, F. W. Holly, J. D. Willet, L. H. Sarett, W. J. Holtz, E. A. Risley, G. W. Nuss, and C. A. Winter, *J. Amer. Chem. Soc.*, 1963, 85, 488.
[49] D. J. Drain, B. Davy, M. Horlington, J. G. B. Howes, J. M. Scruton, and R. A. Selway, *J. Pharm. Pharmacol.*, 1971, 23, 857.
[50] P. Bey, C. Danzin, V. van Dorsselaer, P. Mamont, M. Jung, and C. Tardiff, *J. Med. Chem.*, 1978, 21, 50.
[51] J. G. Atkinson, Y. Giraud, J. Rokach, C. S. Rooney, C. S. McFarlane, A. Rackham, and N. N. Share, *J. Med. Chem.*, 1979, 22, 99.
[52] D. R. Curtis, A. W. Duggan, D. Felix, and G. A. R. Johnston, *Brain Res.*, 1971, 32, 69.
[53] D. R. Curtis and J. C. Watkins, *Nature*, 1961, 191, 1010.

Table 4 *continued*

Prostaglandin ring system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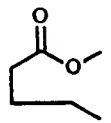
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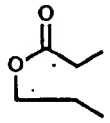
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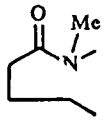
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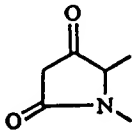
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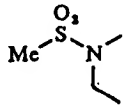
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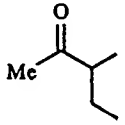
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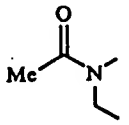
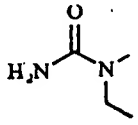
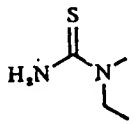
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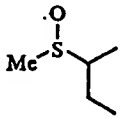
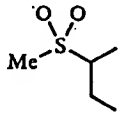
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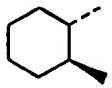
ref. 60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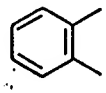
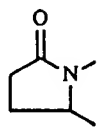


[64] P. A. Zoretic, P. Soja, and T. Shiah, *Prostaglandins*, 1978, 16, 555.
[65] P. A. Zoretic, P. Soja, and T. Shiah, *J. Med. Chem.*, 1978, 21, 1330.
[66] C. J. Harris, N. Whittaker, G. A. Higgs, J. M. Armstrong, and P. M. Reed, *Prostaglandins*, 1978, 16, 773.
[67] J. H. Jones, W. J. Holtz, J. B. Bicking, E. J. Cragoe, R. Mandel, and F. A. Kuehl, *J. Med. Chem.*, 1977, 20, 1299.
[68] J. B. Bicking, C. M. Robb, R. L. Smith, E. J. Cragoe, F. A. Kuehl, and L. R. Mandel, *J. Med. Chem.*, 1977, 20, 35.
[69] J. H. Jones, W. J. Holtz, J. B. Bicking, E. J. Cragoe, L. R. Mandel, and F. A. Kuehl, *J. Med. Chem.*, 1977, 20, 44.
[70] R. L. Smith, J. B. Bicking, N. P. Gould, T.-J. Lee, C. M. Robb, F. A. Kuehl, L. R. Mandel, and E. J. Cragoe, *J. Med. Chem.*, 1977, 20, 540.
[71] T. A. Eggelte, H. de Koning, and H. O. Huisman, *Rec. Trav. chim.*, 1977, 96, 271.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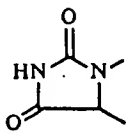
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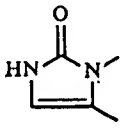
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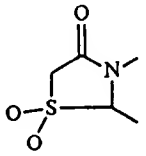
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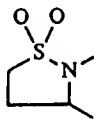
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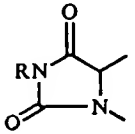
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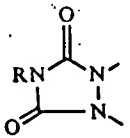
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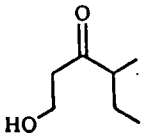
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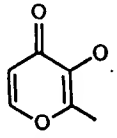
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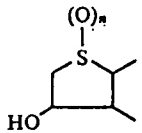
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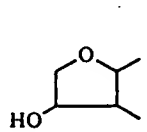
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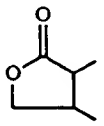
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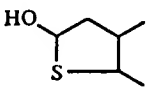
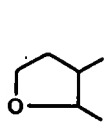

ref. 72

[72] P. A. Zoretic, B. Branchard, and N. D. Sirka, *J. Org. Chem.*, 1977, 42, 3201; J. Bruin, H. de Koning, and H. O. Huisman, *Tetrahedron Letters*, 1975, 4599; G. Bollinger and I. M. Muchowski, *Tetrahedron Letters*, 1975, 2931.
[73] R. L. Smith, T.-J. Lee, N. P. Gould, E. J. Cragoe, H. G. Oien, and F. A. Kuehl, *J. Med. Chem.*, 1977, 20, 1292.
[74] Merck, U.S.P., 4 087 435.
[75] Beechams, Belgian P., 861 956.
[76] Beechams, Belgian P., 861 957.
[77] Miles, U.S.P., 4 127 612.
[78] Pfizer, U.S.P., 4 132 847.
[79] J. Vlattas and L. Dellavecchia, *Tetrahedron Letters*, 1974, 4459.
[70] J. Vlattas and L. Dellavecchia, *Tetrahedron Letters*, 1974, 4455.
[71] Tanabe Seijaku, G.P., 2 229 225; F. M. Hauser and R. C. Huffman, *Tetrahedron Letters*, 1974, 905.
[72] J. T. Harrison, R. J. K. Taylor, and J. H. Fried, *Tetrahedron Letters*, 1975, 1165.

Table 4 *continued*

Prostaglandin ring system (continued)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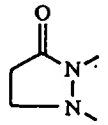
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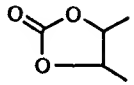
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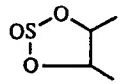
ref. 76

[73] J. T. Harrison, V. R. Fletcher, and J. H. Fried, *Tetrahedron Letters*, 1974, 2733.
[74] E. I. du Pont de Nemours, B.P., 1 428 431.
[75] J. T. Harrison and V. R. Fletcher, *Tetrahedron Letters.*, 1974, 2729.
[76] A. P. Bender, *J. Med. Chem.*, 1975, **18**, 1094.

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